

CASE REPORT

M. Herold · A. Schulze · K. Härtwig · G. Anger

Successful treatment and re-treatment of resistant B-cell chronic lymphocytic leukemia with the monoclonal anti-CD 20 antibody rituximab

Received: 5 May 1999 / Accepted: 18 October 1999

Abstract We report on two patients with chemoresistant B-cell chronic lymphocytic leukemia who were treated successfully with the monoclonal anti-CD 20 antibody rituximab. Both patients suffered from severe thrombocytopenia requiring platelet transfusions over a period of several months. Neither chemotherapy nor immunosuppressive agents (corticoids, immunoglobulins) were effective. After four doses of rituximab (375 mg/m² weekly), both patients recovered within a few weeks to hematological partial remission. One patient was re-treated successfully three times after relapses. Both patients were pre-medicated with prednisone (100 mg) 30 min prior to the infusion to prevent cytokine release and the antibody infusions were well tolerated.

Key words B-cell chronic lymphocytic leukemia · B-CLL · Rituximab · Monoclonal antibody · Immunotherapy

Introduction

The cell surface antigen CD 20 is expressed on more than 90% of B-cell lymphomas, including B-cell chronic lymphocytic leukemia (B-CLL) [2, 9]. In normal B-cells, it is expressed from pre-B-cell stage to the activated B-cell stage, but not on the stem cells or plasma cells. CD 20 is a 37-kDa phosphoprotein composed of 297 amino acids. It is a transmembrane protein acting as a calcium channel and plays an important role in cell-cycle progression [17].

Rituximab (IDEA C2B8) is a chimeric human-mouse monoclonal antibody [15]. It can deplete nor-

mal and malignant B-cells through complement-dependent cytotoxicity, antibody-dependent cell-mediated cytotoxicity [15], and apoptosis induction [7, 16]. Furthermore, in vitro results suggest that the antibody sensitizes drug-resistant cell lines to cytotoxic chemotherapy [8]. Phase-I/II studies of single-agent anti-CD 20 antibody treatment in relapsed B-cell lymphomas have revealed activity in approximately half of the treated patients [11, 12]. Adverse reactions were reported to be mild and were usually related to the first antibody infusion. The results of a multi-center pivotal trial enrolling 166 relapsed patients with low-grade or follicular lymphoma confirmed the phase-I/II results; a response rate of 48%, including 6% complete remissions and a median time to progression of 13 months for the responders [13]. To date, there has been insufficient data regarding the safety and efficacy of rituximab in B-CLL patients. The above-mentioned phase-I/II studies and the pivotal trial excluded patients with lymphocytic lymphoma with leukocyte counts greater than 5 Gpt/l. Recently, several cases of severe tumor lysis syndrome and/or cytokine release after rituximab in B-CLL patients with a high tumor burden have been reported [3, 10]. This corresponds with observations from former trials suggesting that the intensity of infusion-related adverse reactions depends on tumor burden (especially circulating CD 20+ cells) and occurs mainly during the first infusion [11–13].

We report on two chemotherapy-resistant B-CLL patients who were treated successfully and safely with rituximab.

Case reports

Case 1

A 57-year-old female patient with B-CLL stage Binet C (CD 5+, CD 19+, CD 20+, CD 23+) was diagnosed in 1987 by means of lymph-node biopsy. Chemotherapy with bendamustine, vincristine, and prednisolone induced a partial remission, and treat-

M. Herold (✉) · A. Schulze · K. Härtwig · G. Anger
2. Medizinische Klinik, Klinikum Erfurt GmbH, PF 595,
D-99012 Erfurt, Germany
Tel.: +49-361-7813502
Fax: +49-361-7814803
e-mail: mherold@tumorzentrum-erfurt.de

ment was stopped in 1991. As a result of disease progression, chemotherapy with intermittent chlorambucil and prednisolone was started in 1996. Again, a hematological partial remission was achieved.

In December 1997, the patient was admitted to our hospital because of severe anemia, thrombocytopenia (hemoglobin 9.6 g/dl; leukocytes 7.84 Gpt/l with 70% CD 20+ lymphocytes; platelets 24 Gpt/l) and systemic B-symptoms. The clinical examination showed a spleen 4 cm below the costal margin but no lymphadenopathy. Bone-marrow biopsy demonstrated a diffuse lymphatic marrow involvement. Different chemotherapy protocols (bendamustine, vincristine, prednisolone; mitoxanthrone, chlorambucil, prednisolone) failed, as did high-dose immunoglobulins (Igs). During 170 days of hospitalization from December 1997 to July 1998, the patient received 28 U of thrombapheresis concentrates and 21 U of packed red cells. Repeatedly, episodes of fever of unknown origin (FUO) were treated with antibiotics. In June 1998, hemoglobin was 8.0 g/dl, leukocytes 1.9 Gpt/l (61% CD 20+ lymphocytes), platelets 15 Gpt/l, lactate dehydrogenase (LDH) 13.42 µmol/sxl and IgG 2.4 g/l. We decided to start rituximab (375 mg/m² × 4) given in weekly intervals from 12 June 1998 to 2 July 1998. Under premedication with prednisolone (100 mg p.o.), paracetamol (1000 mg p.o.), and dexamethasone (4 mg p.o.) 30 min prior to the 5-h antibody infusion, the patient had no infusion-related adverse reactions. After the third rituximab infusion, the platelets increased to 36 Gpt/l, leukocytes were 2.89 Gpt/l (32% CD 20+ lymphocytes), hemoglobin 9.3 g/dl, and LDH decreased to 7.82 µmol/sxl. The last thrombapheresis concentrate was given on 26 June 1998. Later controls showed increasing platelet counts close to the normal value (September 1998 95 Gpt/l, October 1998 115 Gpt/l, January 1999 100 Gpt/l).

The patient has been followed on an outpatient basis without treatment for nearly 1 year now, showing only slight signs of disease progression. Currently, the hemoglobin level is 9.76 g/dl, leukocytes 6.9 Gpt/l (43% CD 20+ lymphocytes), platelets 56 Gpt/l, LDH 4.66 µmol/sxl, and IgG 2.3 g/l.

Case 2

B-CLL of Binet-stage C was histologically diagnosed in February 1995 in a 63-year-old male patient. The cell surface antigens CD 23, CD 20, CD 19, and CD 5 were expressed on the lymphatic cells. Chemotherapy included chlorambucil/prednisolone; cyclophosphamide, vincristine, prednisolone; mitoxanthrone, chlorambucil, prednisolone; bendamustine and fludarabine, but none of the treatment programs was effective. In November 1997, the patient was admitted to our hospital because of bleeding due to severe thrombocytopenia refractory to platelet substitution, steroids, and immunoglobulins.

Clinical examination showed a generalized lymphadenopathy of 1–2 cm in diameter and an enlargement of liver and spleen of 6 cm below the costal margin. The bone-marrow biopsy revealed a diffuse lymphatic infiltration with nearly complete replacement of the normal hematopoiesis. Hemoglobin was 11.84 g/dl, leukocytes 37.4 Gpt/l (CD 20+ lymphocytes 77%), platelets 9 Gpt/l, LDH 8.55 µmol/sxl, and IgG 9.08 g/l. Rituximab treatment was initiated on 10 November 1997. The antibody was given four times in weekly intervals at a dose of 375 mg/m² as a 5-h infusion. Premedication was paracetamol (1000 mg p.o.) and dexamethasone (4 mg p.o.) 30 min prior to the antibody infusions. Adverse reactions were mild with subfebrile temperatures up to 37.8°C and mild hypertension with a maximal decrease of the systolic blood pressure of 15 mmHg during the infusions. An increase of the platelet count was observed at the end of December 1997 (29 Gpt/l), reaching 152 Gpt/l in January 1998. In February 1998, the patient suffered from mycoplasma pneumonia with a hemolytic episode. Under antibiotic treatment with erythromycin, he recovered within 3 weeks. In April 1998, pulmonary infiltrates were observed and cytomegalovirus was found in serum and bronchoalveolar fluid (using PCR). Gancy-

clovir treatment was given for 8 weeks followed by a complete restitutio of the pulmonary infiltrates. During this period, hematological parameters were stable (hemoglobin 9.5 g/dl, leukocytes 6.5 Gpt/l (50% lymphocytes), platelets 175 Gpt/l, reticulocytes 27%).

In late July 1998, after more than 8 months of freedom from progression, the patient relapsed with a platelet count of 3 Gpt/l and bleeding. A second rituximab treatment started on 1 August 1998. Again four doses of 375 mg/m² were given. Premedication with prednisolone (100 mg i.v.) 30 min prior to the antibody prevented any infusion-related adverse reactions. Recovery of platelets occurred within 5 weeks to a maximum of 90 Gpt/l. However, after 4 months, the patient relapsed again (platelets 6 Gpt/l on 7 December 1998). The third antibody treatment (375 mg/m² × 4 weekly) was started on 14 December 1998. Again, no infusion-related adverse reactions were seen under steroid premedication. The platelet count improved up to a maximum of 121 Gpt/l in April 1999; further clinical and laboratory data: generalized lymphadenopathy of 1 cm in size, spleen 4 cm below the costal margin, hemoglobin 13.6 g/dl, leukocytes 12.3 Gpt/l (30% CD 20+ lymphocytes), LDH 7.75 µmol/sxl. The fourth relapse after now 5 months occurred in May 1999 with signs of bleeding (leukocytes 14.0 Gpt/l with a proportion of 45% CD 20+ lymphocytes, platelets 18 Gpt/l).

Rituximab treatment consisted of only 2 × 375 mg/m² in a weekly interval (12 May to 19 May 1999). Again, there were no infusion-related events under the premedication mentioned above. The latest control on 21 July 1999 showed the patient to be well, with peripheral lymph nodes of less than 1 cm in size, the spleen 3 cm below the costal margin, and hemoglobin 14.24 g/dl, leukocytes 8.1 Gpt/l (45% CD 20+ cells), and platelets 84 Gpt/l.

Discussion

The chimeric monoclonal anti-CD 20 antibody, rituximab, has demonstrated remission rates up to 50% in relapsed low-grade non Hodgkin's lymphomas, especially follicular lymphomas [11, 12, 13]. In combination with CHOP-chemotherapy in mainly untreated patients with low-grade and follicular lymphomas, the response rate reported was 95%. More than half of these patients were complete responders [5]. Since trials reported to date have excluded classical B-cell chronic lymphocytic leukemia, there is no data or experience regarding the potential of rituximab for the treatment of B-CLL. The density of CD 20 expression on most B-CLL cells is lower than in other B-cell lymphomas [18], suggesting B-CLL is less susceptible to anti-CD 20 antibody treatment. However, severe infusion-related toxicity has been reported in several cases of B-CLL and B-PLL treated with rituximab, and similar events occurred in patients treated with rituximab alone or in combination with fludarabine in phase-II clinical trials of the German CLL-study group [3, 10].

Both of the patients reported here were treated effectively and without significant toxicity. In both patients, we had no evidence of immune thrombocytopenic purpura (ITP), which may also respond to rituximab treatment [14]. The first patient with a low number of circulating lymphocytes had no infusion-related adverse reactions after steroid premedication. Response occurred after 3 weeks, and now, after

1 year, there is only mild disease progression that does not require treatment. The second patient had a higher tumor mass at the start of his first antibody treatment with 27.8 Gpt/l of circulating CD 20+ lymphocytes. The first treatment in November 1997 was initiated without steroid premedication, and the patient experienced mild hypotension and subfebrile temperatures. The lymphocyte counts decreased within 24 h to 13.8 Gpt/l. Six weeks after the first rituximab infusion, the patient improved remarkably and was free of progression for 8 months, at which time the first relapse occurred and a second antibody treatment was initiated. As a result of premedication with prednisolone, no infusion-related adverse reactions were seen and the patient responded with hematological partial remission for 4 months and again for 5 months after the third application of rituximab in December 1998. The fourth antibody treatment with only two antibody-doses of 375 mg/m² has again resulted in a response (platelets 84 Gpt/l in July 1999).

In this particular patient, we have demonstrated that re-treatment with rituximab is applicable and effective. This confirms reports of a phase-II trial presented recently [6]. In summary, we regard rituximab to be potentially effective in relapsed and chemotherapy-resistant B-CLL. We consider the severe complications that have been reported, particularly in patients with a high tumor burden, to be mainly related to cytokine release [1, 4] or it may be a combination of different mechanisms including tumor-cell agglutination and destruction followed by cytokine release, as discussed by Byrd et al. [3]. We would recommend steroid premedication (e.g., prednisolone 100 mg i.v.) 30 min prior to the rituximab infusion. This premedication obviously does not impair the efficacy of the antibody as demonstrated in our patients. As a result of a panel discussion during the recent lymphoma conference in Lugano, Switzerland, steroid premedication for rituximab treatment was recommended and has been introduced meanwhile into several clinical trials.

It needs to be stressed that anti-CD 20 antibody treatment of B-CLL is still an experimental treatment approach. All B-CLL patients who are considered for treatment with rituximab should be referred to treatment centers experienced in this modality. A fractionated application of rituximab in the first infusion should be considered, especially in patients with a high tumor burden as well. If possible, all these patients should be included in ongoing clinical phase-II trials.

References

- Alegre ML, Vandenebeele P, Depierreux M, Florquin S, Deschotter-Lanckman M, Flamend V, Moser M, Leo O, Urbain J, Fiers W, Goldman M (1991) Cytokine release syndrome induced by the 145-2C11 anti-CD3 monoclonal antibody in mice: prevention by high doses of methylprednisolone. *J Immunol* 146: 1184-1191
- Anderson KC, Bates MP, Slougenhoupt BL, Pinkus GS, Schlossman SF, Nadler LM (1984) Expression of human B-cell associated antigens on leukemias and lymphomas: a model of human B cell differentiation. *Blood* 63: 1424-1433
- Byrd JC, Waselenko JK, Maneatis TJ, Murphy T, Ward FT, Monahan BP, Sipe MA, Donegan S, White CA (1999) Rituximab therapy in hematologic malignancies patients with circulating blood tumor cells: association with increased infusion related side effects and a rapid tumor clearance. *J Clin Oncol* 17: 791-795
- Chatenoud L, Legendre C, Ferran C, Bach J-F, Kreis H (1991) Corticosteroid inhibition of the OKT3-induced cytokine-related syndrome - dosage and kinetics prerequisites. *Transplantation* 51: 334-338
- Czuczman MS, Grillo-Lopez AJ, White CA, Sahleh M, Gordon L, LoBuglio AF, Jonas C, Klippenstein D, Dallaire B, Varns C (1999) Treatment of patients with low-grade B-cell lymphoma with the combination of chimeric anti-CD 20 monoclonal antibody and CHOP chemotherapy. *J Clin Oncol* 17: 268-276
- Davis T, Levy R, White CA, Czuczman MS, McLaughlin P, Link B, Varns C, Weaver R, Grillo-Lopez AJ (1998) Rituximab: phase II retreatment study in patients with low-grade or follicular NHL (abstract 1710). *Blood* 92[suppl 1]: 419a
- Demidem A, Hanna N, Hariharan H, Bonavida B (1995) Chimeric anti-CD 20 antibody (IDEc C2B8) is apoptotic and sensitizes drug resistant human B-cell lymphoma and AIDS related lymphomas to the cytotoxic effect of CDDP, VP16 and toxic (abstract). *FASEB J* 9: A206
- Demidem A, Lam T, Alias S, Haritharan H, Hanna N, Bonavida B (1997) Chimeric anti-CD 20 antibody sensitizes a B-cell lymphoma cell line to cell killing by cytotoxic drugs. *Cancer Biother Radiopharm* 12: 177-186
- Harris NL, Jaffe ES, Stein H, Banks PM, Chan JKC, Cleary ML, Delsol G, DeWolf-Peeters C, Falini B, Gatter KC, Grongan TM, Isaacson PG, Knowles DM, Mason DY, Müller-Hermelink HK, Pilieri SA, Piris MA, Ralfkiaer Wamke RA (1994) A revised European-American classification of lymphoid neoplasms: a proposal from the International Lymphoma Study Group. *Blood* 84: 1361-1392
- Jensen M, Winkler U, Manzke O, Diehl V, Engert A (1998) Rapid tumor lysis in a patient with B-cell chronic lymphocytic leukemia and lymphocytosis treated with an anti-CD 20 monoclonal antibody (IDEc C2B8, rituximab). *Ann Hematol* 77: 89-91
- Maloney D, Liles T, Czerwinski D, Walden KC, Rosenberg J, Grillo-Lopez AJ, Levy R (1994) Phase I clinical trial using escalating single-dose infusion of chimeric anti-CD 20 antibody (IDEc C2B8) in patients with recurrent B-cell lymphomas. *Blood* 84: 2457-2466
- Maloney D, Grillo-Lopez AJ, White CA, Bodkin D, Schilder RJ, Neidhardt JA, Janakiraman N, Foon KA, Liles TM, Dallaire BK, Wey K, Royston I, Davis T, Levy R (1997) IDEC C2B8(Rituxan AB) anti-CD 20 monoclonal antibody therapy in patients with relapsed low-grade non-Hodgkin's lymphomas. *Blood* 90: 2188-2195
- McLaughlin P, Grillo-Lopez AJ, Link BK, Levy R, Czuczman MS, Williams ME, Heyman MR, Bluce-Buckler I, White CA, Cabanillas F, Jain V, Ho AD, Lister J, Wey K, Shen D, Dallaire BK (1998) Rituximab chimeric anti-CD 20 monoclonal antibody therapy for relapsed indolent lymphoma: half of patients respond to a four-dose treatment program. *J Clin Oncol* 16: 2825-2833

14. Perrotta A, Abuel C (1998) Response of chronic relapsing ITP of 10 years duration to rituximab (abstract 3360). *Blood* 92(suppl 1):88b
15. Reff ME, Carner K, Chambers KS, Chinn PC, Leonard JE, Raab R, Newman RA, Hanna N, Anderson DR (1994) Depletion of B cells in vivo by a chimeric mouse human monoclonal antibody to CD 20. *Blood* 83:435-445
16. Riethmüller G, Schneider-Gädicke E, Johnson JP (1993) Monoclonal antibodies in cancer therapy. *Curr Opin Immunol* 5:732-739
17. Tedder TF, Engel P (1994) CD 20: a regulator of cell-cycle progression of B lymphocytes. *Immunol Today* 15:450-454
18. Tefferi A, Bartholmai BJ, Witzig TE, Li CY, Hauson CA, Phylky RL (1996) Heterogeneity and clinical relevance of the intensity of CD 20 and immunoglobulin light-chain expression in B-cell chronic lymphocytic leukemia. *Am J Clin Pathol* 106:457-461